A Phase I Vaccine Safety and Chemotherapy Dose-Finding Trial of an Allogeneic GMCSF-secreting Breast Cancer Vaccine Given in a Specifically Timed Sequence with Immunomodulatory Doses of Cyclophosphamide and Doxorubicin

Nontechnical Abstract

Breast cancer is the most common cancer and the second leading cause of cancer death in women. In the year 2004, there will be an estimated 217,440 new cases of breast cancer diagnosed and approximately 40,580 deaths due to the disease. While 80% of patients present with locoregional disease involving the breast and/or axillary lymph nodes, about half develop disseminated disease and ultimately die from it. Stage 4 breast cancer is typically managed with hormonal agents or conventional cytotoxic drugs. Tumors quickly become resistant to these treatments, however, underscoring the need for novel therapeutic strategies that can be integrated with existing therapeutic modalities in an additive or synergistic fashion. Immunotherapy is a particularly attractive strategy for overcoming drug resistance. Immunotherapy is a type of treatment for cancer based on the idea that the immune system can be activated to destroy cancer cells that might be resistant to hormonal therapy and chemotherapy. A vaccine is a kind of immunotherapy that delivers an antigen (something that activates the immune system) so that the immune system recognizes cells with that antigen as foreign and destroys any cells that display that antigen.

The allogeneic breast tumor cell vaccine consists of two types of breast tumor cells developed from the tumor cells of patients with breast cancer. The human granulocyte-macrophage colony-stimulating factor (GM-CSF) gene was used to genetically modify the breast tumor cells to secrete GM-CSF. GM-CSF is a substance made by the body that helps the immune system recognize a tumor and destroy it. The vaccine cells were irradiated to prevent them from growing or dividing. The cells themselves are **not** radioactive. The cells are stored frozen until the day of vaccination. The total number of cells in each vaccination for twenty seven people will be 500,000,000, divided into twelve injections given in the thighs and arms. The choice of twelve injections for each vaccination is based on the volume of the vaccination and a finding that the body has a better chance to respond to the vaccine if it is injected into a number of different areas. Because this particular vaccine has not been tested in people before, the first three people will get a vaccination with a total of 50,000,000 cells divided into three injections, one in each thigh and one in one arm.

We propose to test the safety and bioactivity of an allogeneic GM-CSF-secreting breast cancer vaccine when given alone, and when given in a specifically timed sequence with Cyclophosphamide (CY) and Doxorubicin (DOX), two drugs commonly used to treat breast cancer. In this study the CY and DOX are used at lower doses than usual to help the vaccine to activate the patient's immune system. The doses and scheduling of CY and DOX used are based on testing the drugs with a GM-CSF-secreting vaccine in mice that get breast cancer, and are the ones that enabled the vaccine to induce the most potent anti-tumor immunity. The doses of vaccine cells are based on the safety of the same doses of a similar GM-CSF-secreting vaccine for pancreatic cancer. The first six people will get vaccine cells alone, three at a dose of 50,000,000 cells and three at a dose of 500,000,000 cells. The next twenty four people will receive a fixed dose of 500,000,000 allogeneic breast tumor vaccine cells containing two

irradiated allogeneic breast cancer cell lines transfected with the GM-CSF gene in a specifically timed sequence with a range of low doses of CY and DOX. Patients will receive CY intravenously on Day -1, vaccination on Day 0, and DOX intravenously on Day +7. Patients will receive three monthly vaccination cycles, with a fourth and final (boost) vaccination cycle three months from the third cycle.

Blood samples to measure GM-CSF levels will be taken on the day of vaccination and then every day for 4 days. Blood samples to evaluate the safety of the vaccinations will be taken once a week for one month following each vaccination. Blood samples may be taken near the patient's home and sent to Johns Hopkins for testing. During studies of similar vaccines in renal cell cancer, prostate cancer, pancreatic cancer, and non-small cell lung cancer, local symptoms of swelling and redness developed at the vaccine site between 2 and 7 days after vaccination. In this study, if the patient's vaccination site shows swelling over 1 cm in diameter, a skin biopsy will be taken. The skin biopsy will be evaluated to determine to what types of cells are important to the immune response. Based on our previous preclinical and clinical data, the biopsy will be taken on day 3, and possibly on day 7, after vaccination. Other tests and evaluations include history and physical examination, vital signs, CT of the chest, abdomen, and pelvis, nuclear medicine bone scan, pre-vaccination biopsy, blood for immune monitoring, and a skin test for delayed-type hypersensitivity (DTH) that is like a PPD test and involves injecting pieces of a protein antigen *(HER-2/neu)* that is delivered by the breast cancer vaccine. The purpose of the DTH test is to evaluate whether the patient has developed a systemic immune response to the breast cancer vaccine.

This study is a Phase I trial testing the safety and bioactivity of two lethally irradiated GM-CSF-transfected allogeneic breast cancer cell lines injected intradermally either alone, or in a specifically timed sequence with a range of low doses of Cyclophosphamide (CY) and Doxorubicin (DOX) in women with metastatic breast cancer. We began recruiting research subjects into the study on January 13, 2004 with presentation of the consent form to the first interested patient, and the study was initiated on January 15, 2004 when the subject gave informed consent. Since that time, we have consented a total of two research subjects on study, both of whom met eligibility criteria. The first research subject received the first vaccination on February 2, 2004, and has now completed two vaccination cycles. The second research subject has been vaccinated once (March 22, 2004). Both study subjects have experienced local vaccine site reactions including redness, swelling, and itching.

There were no serious adverse events during this reporting period. However, our Institutional Review Board has instituted a new category of adverse event termed an important adverse event. This is defined as an event, although not a serious adverse event, that presents an undesirable occurrence that interferes with the research subject's usual activities and may be persistent or require treatment. One research subject developed abdominal pain nine days after receiving the second vaccination. This pain required evaluation in the outpatient clinic, and the research subject received a dose of narcotic for pain control, and was sent home on Toradol. That same evening the pain spontaneously resolved two days after it began. This important adverse event was deemed probably not related to the vaccine therapy.